(b) subjecting the operator DNA sequence to a mutagenesis, and

(c) analyzing the operator DNA sequences to determine whether said sequences have a different thermostability as compared to a wild-type sequence with regard to binding a repressor.

Div. cont.

39. The method according to claim 38, wherein the lambdoid phages are selected from the group consisting phage lambda, phage 21, phage 22, phage 82, phage 424, phage 434, phage D326, DLP12, phage gamma, phage HKO22, phage P4, phage Phi80, phage Phi81, and coliphage 186.

40. The method according to claim 39, wherein said lambdoid phage is phage lambda.

B1 Cont'

41. The method according to claim 40, wherein said operator DNA sequence is from the operator regions OR and/or OL of the phage lambda.

42. The method according to claim 38, wherein said selection gene is an E-lysis gene from phage PhiX174.

43. The method according to claim 38, wherein the operator DNA sequence is subjected to a site-specific mutagenesis by oligonucleotides or a selection is carried out in a mutator bacterial strain.

44. The method according to claim 38, wherein the operator DNA sequences are analyzed by determining their ability to bind to a temperature-sensitive cI repressor.

45. The method according to claim 44, wherein said temperature-sensitive lambda cI repressor is cI857.

46. An OR or OL operator sequence from lambdoid phages which have an increased thermostability compared to a wild-type sequence with regard to binding of a temperature-sensitive cI repressor, wherein said sequences are obtained by a method comprising

(a) preparing a DNA cassette which contains a selection gene under the operative control of an expression control sequence comprising at least one OR or OL operator DNA sequence from a lambdoid phage and a promoter,

(b) subjecting the operator DNA sequence to a mutagenesis, and

(c) analyzing the operator DNA sequences to determine whether said sequences have an

increased thermostability as compared to a wild-type sequence with regard to binding a repressor.

47. The OR or OL operator sequence according to claim 46, wherein it has an approximately 3 - 10°C increased thermostability.

48. The OR or OL operator sequence according to claim 47, wherein it has an approximately 7 - 9°C increased thermostability.

49. A lambda OR operator sequence comprising the sequence shown in SEQ ID NO. 2.

50. A nucleic acid comprising a bacterial expression control sequence containing a OR or OL operator sequence according to claim 46 in operative linkage with a protein-coding sequence.

51. The nucleic acid according to claim 50, wherein the protein-coding sequence is a suicide gene.

52. The nucleic acid according to claim 53, wherein the expression control sequence contains a lambda PL or PR promoter.

53. A vector comprising at least one copy of a nucleic acid according to claim 50.

54. The vector according to claim 53, wherein said vector is a bacterial chromosomal vector.

55. The vector according to claim 53, wherein said vector is a bacterial extrachromosomal plasmid.

56. A bacterial cell transformed with a nucleic acid according to claim 50.

57. A bacterial cell transformed with a vector according to claim 53.

58. A bacterial cell according to claim 56, wherein said nucleic acid is integrated into said cell's chromosome.

B1 Cont.

59. A bacterial cell according to claim 57, wherein said vector is integrated into said cell's chromosome.

60. A bacterial cell according to claim 56, further comprising a gene for a cI repressor from lambdoid phages.

61. A bacterial cell according to claim 57, further comprising a gene for a cI repressor from lambdoid phages.

62. A bacterial cell according to claim 60, wherein said gene is the lambda cI857 repressor.

63. A vaccine composition, comprising a live bacterial cell according to claim 56 in combination with pharmaceutically acceptable auxiliary substances, additives or carrier substances.

64. A vaccine composition, comprising a live bacterial cell according to claim 57 in combination with pharmaceutically acceptable auxiliary substances, additives or carrier substances.

65. A vaccine composition, comprising a bacterial ghost in combination with pharmaceutically acceptable auxiliary substances, additives and carrier substances in which the bacterial ghost can be obtained by culturing a bacterial cell as claimed in claim 57 at temperatures of 35 - 39°C and subsequently lysing the bacterial cell by increasing the temperature.

B1 cont.

66. A nucleic acid comprising (a) a first bacterial expression control sequence which contains an OR or OL operator sequence from a lambdoid phage and to which a first cI repressor from lambdoid phages can bind, in operative linkage with a sequence coding for a second repressor wherein the second repressor cannot bind to the first bacterial expression sequence and (b) a second bacterial expression control sequence to which the second repressor can bind in operative linkage with a suicide gene.

67. A bacterial cell, comprising at least one copy of a nucleic acid according to claim 66.

68. The bacterial cell according to claim 67, further comprising a gene for said first cI repressor.

69. The bacterial cell according to claim 67, wherein said first bacterial expression control sequence is an operator sequence from a lambdoid phage wherein said sequence has a different thermostability compared to a wild-type sequence with regard to binding of a repressor and wherein said operator sequence is obtained by a method comprising

(a) preparing a DNA cassette which contains a selection gene under the operative control of an expression control sequence comprising at least one OR or OL operator DNA sequence from a lambdoid phage and a promoter,

(b) subjecting the operator DNA sequence to a mutagenesis, and

(c) analyzing the operator DNA sequences to determine whether said sequences have a different thermostability as compared to a wild-type sequence with regard to binding a repressor.

70. The bacterial cell according to claim 67, further comprising (c) a third bacterial expression control sequence which contains a operator sequence in operative linkage with a suicide gene, wherein said operator sequence is from a lambdoid phage and wherein said operator sequence has a different thermostability compared to a wild-type sequence with regard to binding of a repressor and wherein said operator sequence is obtained by a method comprising

(a) preparing a DNA cassette which contains a selection gene under the operative control of an expression control sequence comprising at least one OR or OL operator DNA sequence from a lambdoid phage and a promoter,

(b) subjecting the operator DNA sequence to a mutagenesis, and

(c) analyzing the operator DNA sequences to determine whether said sequences have a different thermostability as compared to a wild-type sequence with regard to binding a repressor.

71. A vaccine composition, comprising a live bacterial cell according to claim 67 in combination with pharmaceutically acceptable auxiliary substances, additives or carrier substances.

72. The vaccine according to claim 71, wherein said vaccine is a heat-sensitive, a cold-sensitive or both a heat and cold sensitive safe live vaccine.

73. A method for the temperature regulated expression of genes in bacterial cells, comprising transforming a bacterial cell with a nucleic acid comprising a bacterial expression control sequence containing a OR or OL operator sequence according to claim 46 in operative linkage with a protein-coding sequence, and adjusting the temperature to control the expression of said protein-coding sequence.

74. A method for the temperature-regulated sequential expression of genes, comprising transforming a bacterial cell with a nucleic acid comprising (a) a wild-type OR or OL operator region and at least one operator region which contains an OR or OL operator sequence according to claim 46 or (b) several operator regions which contain OR or OL operator sequences according to claim 46 wherein said operator regions have different thermostabilities, and adjusting the temperature to regulate the sequential expression of genes.

B1 cont.

75. The method according to claim 73, wherein said repressor is a cI repressor from a lambdoid phage.

76. The method according to claim 75, wherein said repressor is the lambda cI857 repressor.--